abcam

Product datasheet

Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker ab40839

敲除验证 重组 RabMAb

★★★★★ 5 Abreviews 16 References 17 图像

概述

产品名称	Anti-Ezrin抗体[EP886Y] - Plasma膜Marker
描述	兔单克隆抗体[EP886Y] to Ezrin - Plasma膜Marker
宿主	Rabbit
经测试应用	**适用于:** ICC/IF, Flow Cyt (Intra), WB, IP, IHC-P
种属反应性	**与反应:** Human
免疫原	Synthetic peptide within Human Ezrin aa 450-550 (C terminal). The exact sequence is proprietary.
阳性对照	WB: Wild-type HAP1 whole cell lysate; HeLa and HCT116 whole cell lysates. IHC-P: Human Colon, breast and bladder carcinoma. ICC/IF: HeLa cells Flow Cyt (intra): HeLa cells IP: HeLa cells
常规说明	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol, 0.05% BSA

纯度	Protein A purified
克隆	单克隆
克隆编号	EP886Y
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab40839于以下的经测试应用**

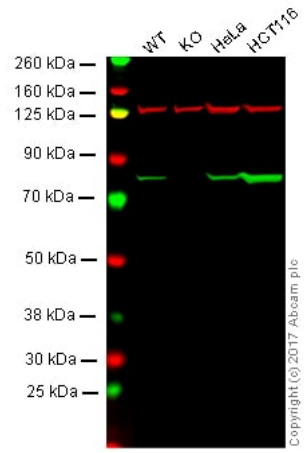
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
ICC/IF	★★★★★ (2)	1/500. **For upurified use at 1/50 - 1/100**
Flow Cyt (Intra)		1/800. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. For upurified use at 1/1000
WB	★★★★★ (2)	1/5000 - 1/50000. Detects a band of approximately 72 kDa (predicted molecular weight: 69 kDa).
IP		1/40. **For upurified use at 1/50**
IHC-P	★★★★☆ (1)	1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.

靶标

功能	Probably involved in connections of major cytoskeletal structures to the plasma membrane. In epithelial cells, required for the formation of microvilli and membrane ruffles on the apical pole. Along with PLEKHG6, required for normal macropinocytosis.
组织特异性	Expressed in cerebral cortex, basal ganglia, hippocampus, hypophysis, and optic nerve. Weakly expressed in brain stem and diencephalon. Stronger expression was detected in gray matter of frontal lobe compared to white matter (at protein level). Component of the microvilli of intestinal epithelial cells. Preferentially expressed in astrocytes of hippocampus, frontal cortex, thalamus, parahippocampal cortex, amygdala, insula, and corpus callosum. Not detected in neurons in most tissues studied.
序列相似性	Contains 1 FERM domain.
发展阶段	Very strong staining is detected in the Purkinje cell layer and in part of the molecular layer of the infant brain compared to adult brain.
翻译后修饰	Phosphorylated by tyrosine-protein kinases.
细胞定位	Apical cell membrane. Cell projection. Cell projection > microvillus membrane. Cell projection > ruffle membrane. Cytoplasm > cell cortex. Cytoplasm > cytoskeleton. Localization to the apical membrane of parietal cells depends on the interaction with MPP5. Localizes to cell extensions and peripheral processes of astrocytes (By similarity). Microvillar peripheral membrane protein.

图片

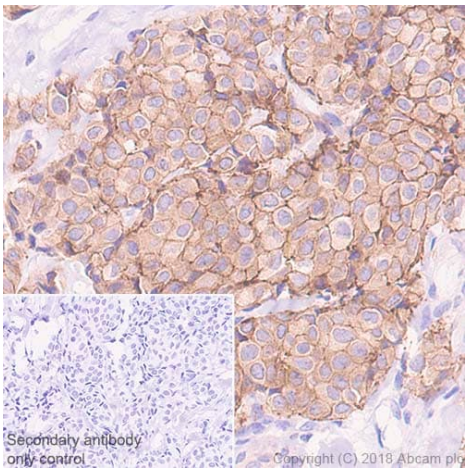


Western blot - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: Ezrin knockout HAP1 whole cell lysate (20 µg)
Lane 3: HeLa whole cell lysate (20 µg)
Lane 4: HCT116 whole cell lysate (20 µg)

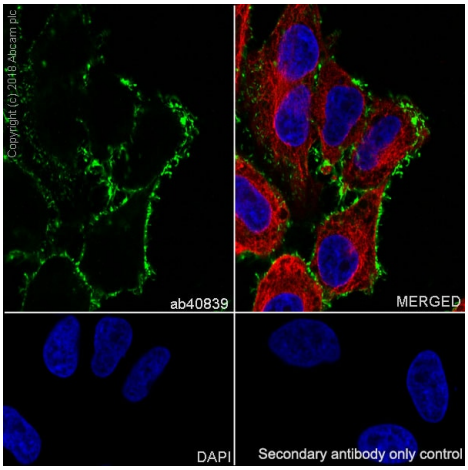
Lanes 1 - 4: Merged signal (red and green). Green - ab40839 observed at 75 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

ab40839 was shown to specifically react with Ezrin in wild-type HAP1 cells as signal was lost in Ezrin knockout cells. Wild-type and Ezrin knockout samples were subjected to SDS-PAGE. Ab40839 and **ab18058** (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/5000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



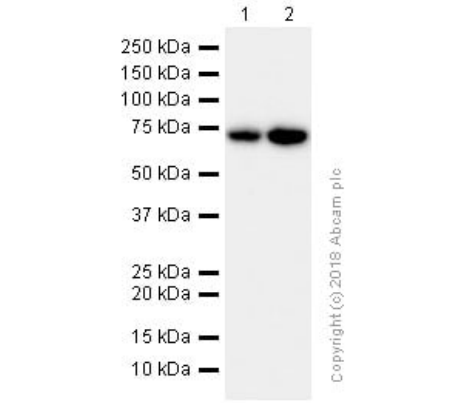
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast carcinoma tissue sections labeling Ezrin with Purified ab40839 at 1:250 dilution (3.28 µg/ml). Heat mediated antigen retrieval was performed Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunocytochemistry/ Immunofluorescence - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Ezrin with Purified ab40839 at 1:500 dilution (1.6 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Western blot - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

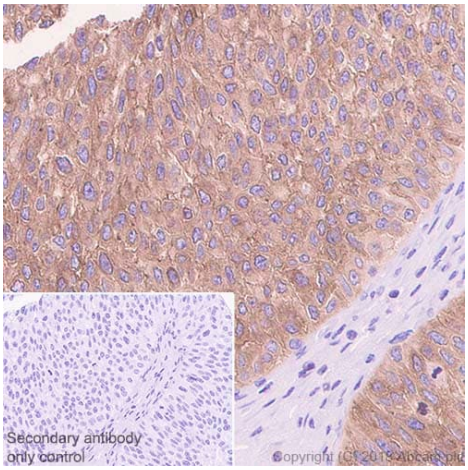
All lanes : Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839) at 1/20000 dilution (Purified)

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates
Lane 2 : HCT 116 (Human colorectal carcinoma epithelial cell) whole cell lysates

Lysates/proteins at 20 µg per lane.

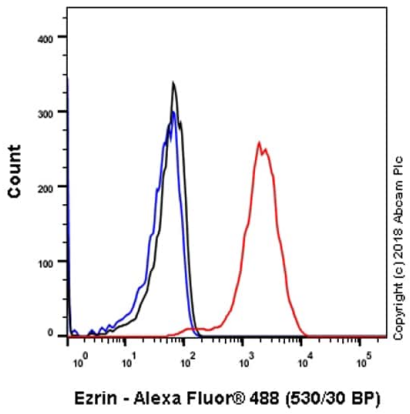
Secondary
All lanes : Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 69 kDa
Observed band size: 72 kDa



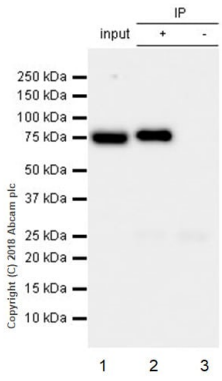
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human bladder carcinoma tissue sections labeling Ezrin with Purified ab40839 at 1:250 dilution (3.28 µg/ml). Heat mediated antigen retrieval was performed Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



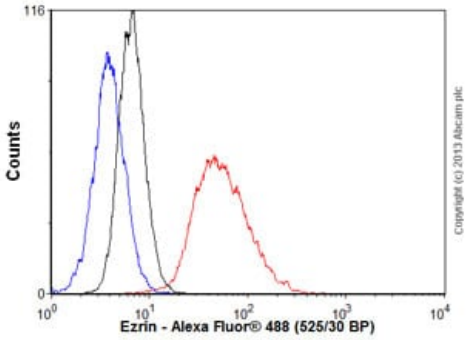
Flow Cytometry (Intracellular) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Ezrin with Purified ab40839 at 1/800 dilution (1 µg/ml) (red). Cells were fixed with 4% Paraformaldehyde. A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



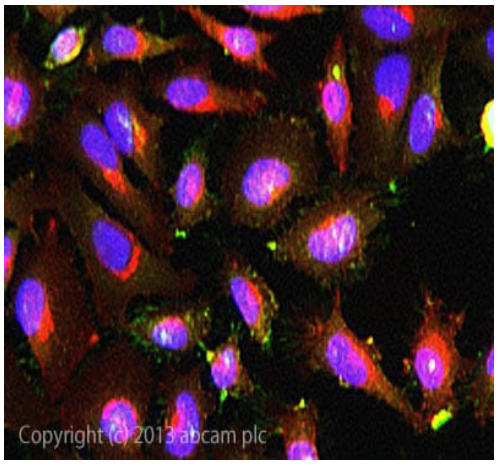
Immunoprecipitation - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

ab40839 (purified) at 1:40 dilution (2µg) immunoprecipitating Ezrin in HeLa whole cell lysate.
Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10µg
Lane 2 (+): ab40839 & HeLa whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of ab40839 in HeLa whole cell lysate
For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used for detection at 1:1000 dilution.
Blocking and diluting buffer: 5% NFDM/TBST.



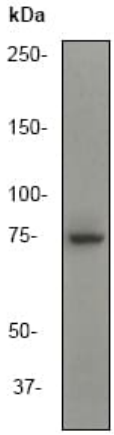
Flow Cytometry (Intracellular) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Overlay histogram showing SH-SY5Y cells stained with upurifiedab40839 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab40839, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in SH-SY5Y cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Immunocytochemistry/ Immunofluorescence - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

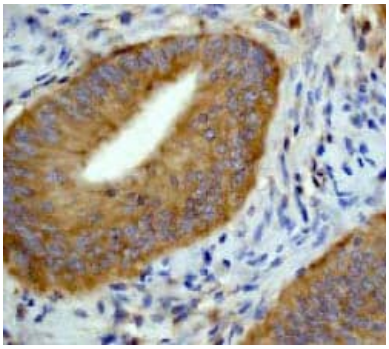
ICC/IF image of unpurified ab40839 stained HeLa cells. The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab40839 at 1/100 dilution overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti- rabbit (**ab96899**) IgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Western blot - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

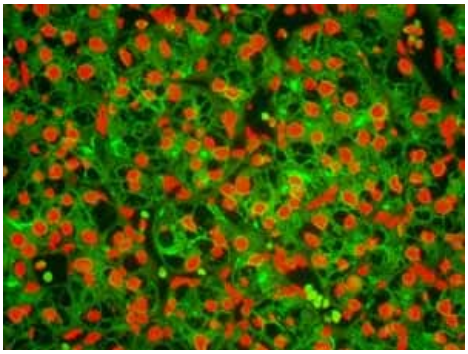
Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839) at 1/50000 dilution (Unpurified) + Hela cell lysate at 10 µg

Predicted band size: 69 kDa
Observed band size: 72 kDa



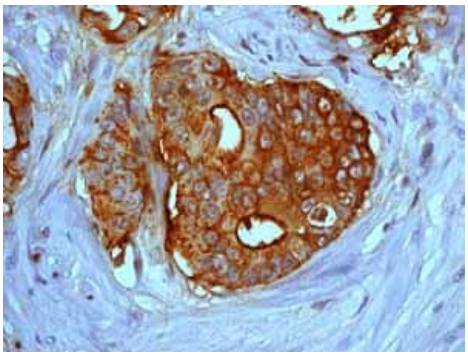
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Unpurified ab40839 at a 1:100 dilution staining Ezrin in human colon carcinoma tissue.



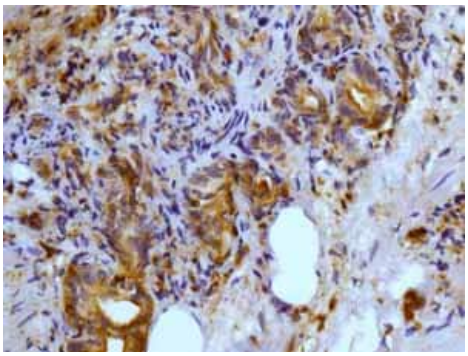
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Fluorescent immunohistochemical analysis of paraffin-embedded human kidney carcinoma tissue using unpurified ab40839. Green-Ezrin red-PI



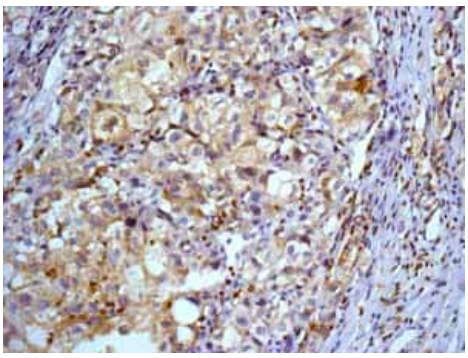
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Unpurified ab40839 showing positive staining in Breast carcinoma tissue.



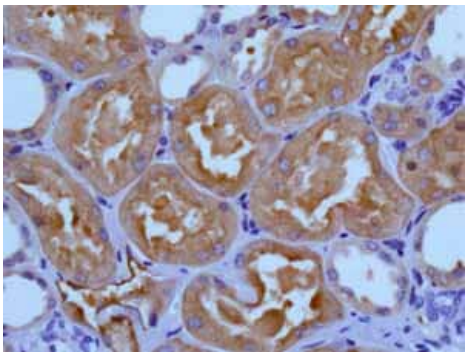
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Unpurified ab40839 showing positive staining in Prostatic carcinoma tissue.



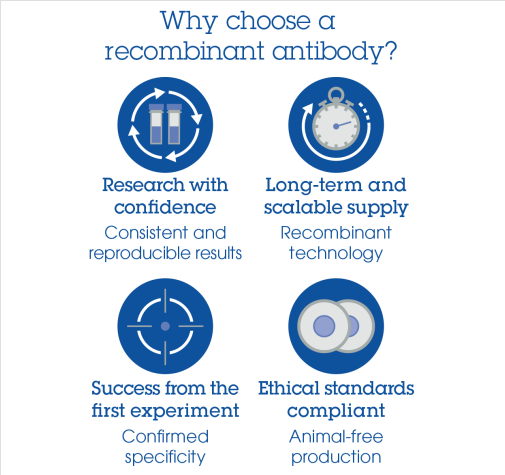
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Unpurified ab40839 showing positive staining in Hepatocellular carcinoma tissue.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

Unpurified ab40839 showing positive staining in Normal kidney tissue.



Anti-Ezrin antibody [EP886Y] - Plasma Membrane Marker (ab40839)

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